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Evaluation of Diffusing Capacity in Patients With a Restrictive Lung Disease*

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Background: **In healthy volunteers, the single-breath diffusing capacity of the lung for carbon monoxide (DLCO) decreases and DLCO normalized per liter alveolar volume (VA; DLCO/VA) increases if VA is decreased. We hypothesized that comparison of DLCO/VA with its predicted value at predicted total lung capacity (TLC) will result in an underestimation of the diffusion disorder in patients with a restrictive lung disease, if a similar relationship exists between DLCO/VA and lung volume as found in healthy volunteers.**

Objective: **To test this hypothesis, we studied total gas transfer DLCO and DLCO/VA as functions of VA in patients who developed a restrictive lung disease and a diffusion disorder in a short period of time.**

Design: **An observational survey.**

Setting: **Pulmonary function department.**

Patients: **Thirteen patients without any initial pulmonary pathology who developed the mentioned pulmonary pathology due to bleomycin treatment.**

Interventions: **Bleomycin treatment.**

Measurements and results: **We performed the single-breath test at various VA levels before, during, and after bleomycin treatment. In the majority of the patients, the DLCO vs VA relationship remained parabolic, but shifted downwards during therapy. Therefore, the linear DLCO/VA vs VA relationship shifted downwards, while the negative slope was not changed, indicating the development of a decreased gas transfer. Six patients also developed a volume restriction.**

Conclusions: **The agreement of the data with the hypothesis increased its probability. Consequently, to evaluate a diffusion disorder, DLCO/VA at a lower actual TLC of patients with a lung restriction should be compared to a reference DLCO/VA at a lung volume equal to the actual TLC.**

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Key words: alveolar volume; bleomycin; chemotherapy; diffusing capacity; restrictive lung disease

Abbreviations: DLCO = diffusing capacity of the lung for carbon monoxide; DLCO/VA = diffusing capacity of the lung for carbon monoxide normalized per liter alveolar volume; Hb = hemoglobin concentration; TLC = total lung capacity; TLCsb = total lung capacity determined with the single-breath test; TLCmb = total lung capacity determined with the multiple-breath He washin method; VA = alveolar volume; VC = vital capacity

In healthy volunteers, the diffusing capacity of the lung for carbon monoxide (DLCO) decreases and the DLCO per liter alveolar volume (VA; DLCO/VA) increases if VA is decreased.[1–12] DLCO/VA vs VA yields a linear relationship with a negative slope.[1,2,11,12] In patients with a volume restriction due to intra- or extraparenchymal diseases, DLCO and DLCO/VA are determined at a total lung capacity (TLC) below their predicted TLC.[13] Based on our previous analyses in healthy volunteers, we advised comparing DLCO/VA in patients with a restrictive lung disease with a reference value counting for a lung volume equal to the patients' TLC.[2,3] However, such advice only holds good if the relationship between DLCO/VA and VA is similar to that in normal subjects. Therefore, we hypothesized that in patients with a restrictive lung disease, DLCO/VA is decreased by the diffusion disorder and increased by the volume decrease. As a consequence, DLCO/VA should be compared with predicted values of DLCO/VA at a lung volume equal to the disease-limited TLC to evaluate the diffusion disorder at alveolar to capillary membrane.

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To test this hypothesis, we studied the volume dependence of the diffusion indexes in a group of patients who gradually developed a diffusion disorder, whether or not in combination with a volume restriction, in a relatively short period. Such processes may occur in patients receiving bleomycin in a chemotherapeutic regimen. An important side effect of bleomycin is lung damage, characterized by pneumonitis or diffuse interstitial pulmonary fibrosis,[14,15] with a decrease in TLC, diffusion indexes, or both.[16] DLCO appeared to be the best indicator of early lung damage.[17] This index enables an early discontinuation of bleomycin treatment at a stage when lung toxicity is still reversible.[18] In a group of patients receiving bleomycin, we estimated DLCO and DLCO/VA at different lung volumes before, during, and after treatment.

MATERIALS AND METHODS

The protocol was approved by the Erasmus University review board for human studies.

Subjects

In 13 adult men suffering from germ cell tumors but without any initial pulmonary pathology, DLCO and DLCO/VA were determined. The spirometric data before the chemotherapeutic treatment, expressed as a mean percentage of predicted values ± SD of the European Community for Coal and Steel,[13] were as follows: TLC, 101 ± 8%; vital capacity (VC), 99 ± 12%; and FEV_1 as a fraction of VC, 98 ± 10%. Mean DLCO and DLCO/VA at TLC, corrected to a normal hemoglobin (Hb) concentration, were 87 ± 14% and 94 ± 15% of the predicted value, respectively.[1] Their ages ranged from 20 to 35 years. Predicted values of Hb concentration were 148 ± 8 g/L and 132 ± 8 g/L in men and women, respectively, as determined in a group of 120 volunteers with the same demographic background in the Laboratory for Clinical Chemistry in our hospital (J. Lindemans, PhD; unpublished data; July 1992).

Procedures and Protocol

In a series of 12 single-breath maneuvers, in which the subjects expired to residual volume and then held their breath after inspiring volumes ranging from 1.5 L up to VC in a random order, DLCO and DLCO/VA were determined in a sitting position at various VA levels.[1] The single-breath procedures were performed with a Masterlab Transfer (Jaeger; Würzburg, Germany) following the European Community for Coal and Steel guidelines.[13] The start of effective breath-holding time was taken when 30% of the inspiration time had elapsed, and the end when half of the expired sample had been collected. Overall breath-holding time slightly exceeded 10 s. Inspirations and expirations were performed rapidly.

Alveolar fractions of CO and He were obtained from expired gas after discarding 800 mL for washout of airways and apparatus dead space. The size of the alveolar sample was 800 mL. At least 5 min elapsed between consecutive measurements. To minimize the influence of CO back tension, the number of measurements was restricted to six a day. Before each series of measurements, patients rebreathed for several minutes in an air-filled bellows system, in which CO_2 was absorbed and O_2 was supplemented. The CO concentration was read from the analyzer, when its level in the rebreathing system was constant. Because this CO concentration in the rebreathing system appeared to be < 1% of the alveolar CO tension at the start of breath holding, back tension was neglected. If a sigh occurred, we waited 5 min before performing the next measurement.[1,5,12,19] The Masterlab Transfer used a heat conductivity type He analyzer that is sensitive to CO_2. Therefore, CO_2 was absorbed prior to both He and CO analysis. The expiratory gas concentrations were corrected for an absorbed volume corresponding to 5% CO_2.[20]

Effects of variation in Hb concentration during the period of chemotherapy were eliminated by correction to the patients' Hb concentration before treatment. For this correction, we used the following equation:

$$\text{DLCO/VA (corr)} = \text{DLCO/VA (obs)}.(a + \theta s[Hb])/([a + \theta s].[Hb])$$

Where DLCO/VA(corr) is DLCO/VA corrected to pretreatment Hb concentration; DLCO/VA (obs) is the observed DLCO/VA at the actual Hb concentration during chemotherapy; a is the ratio of membrane conductance and capillary blood volume (in traditional units [milliliters, minute, and millimeters Hg], approximately 0.7); and θs is the reaction rate for the CO Hb reaction at an O_2 pressure of 110 mm Hg and normal Hb concentration (in traditional units, 1.0).[13,21]

Spirometry, performed with a water-sealed spirometer, and the DLCO/VA vs VA relationships were determined before treatment and immediately after two and four chemotherapeutic treatments. The measurements were repeated at 6 months and at 1 year after the last treatment. Ventilation distribution was evaluated on the basis of the ratio between TLC determined with the single-breath test (TLCsb) and TLC determined with the multiple-breath He washin method (TLCmb). A TLCsb/TLCmb ratio > 0.85 has been proposed as an indication for normal ventilation distribution.[22]

The patients received combination chemotherapy, consisting of cisplatin, 20 mg/m^2 body surface area, on days 1 to 5; etoposide, 100 mg/m^2 body surface area, on days 1 to 5; and 30 mg bleomycin IV push on days 2, 9, and 16. Courses were repeated every 3 weeks. The maximum total dose of bleomycin was 360 mg.

Statistics

Changes were tested with use of a paired Student's *t* test. Differences between two groups of data were regarded as significant at p value < 0.05.

RESULTS

In all patients, the Hb concentration decreased significantly during therapy (Tables 1, 2; p value < 0.001, paired *t* test). After two and four bleomycin treatments, the average decreases in Hb concentration ± SD were 17 ± 8% and 27 ± 8%, respectively. We separated the patients in two groups: in one group, the TLC decreased by > 10% of the pretreatment TLC (Table 1); in the other group, the TLC changed < 10% from baseline TLC (Table 2). Ventilation distribution was evaluated on the basis of the TLCsb/TLCmb ratio. This ratio was > 0.85 in all patients at all stages of therapy.

Table 1—*Change in Hb Concentration, TLC, and DLCO/VA vs VA Relationship Dependent on the Stage of the Chemotherapy in the Patients Who Developed a Restrictive Ventilatory Defect

Pt	After Bleomycin Treatment and Recovery	Hb, g/L	TLC, L	DLCO/VA = a − b VA			DLCO/VA, mL CO/min/mm Hg/L (% Pretreatment)	
				b	a	r	At Actual TLC	At Similar Lung Volume†
1	Pre	154	8.00	0.70	10.78	−0.98	5.17 (100)	6.09 (100)
	2t	140	7.77	0.79	10.80	−0.96	4.64 (90)	5.48 (90)
	4t	129	6.71	0.77	9.64	−0.98	4.48 (87)	4.48 (74)
	1y	150	7.35	0.76	10.07	−0.98	4.48 (87)	4.96 (81)
2	Pre	158	7.56	0.41	7.20	−0.99	4.08 (100)	4.40 (100)
	2t	132	7.91	0.49	7.40	−0.99	3.49 (86)	4.06 (92)
	4t	90	6.78	0.41	6.20	−0.99	3.40 (83)	3.40 (77)
3	Pre	153	7.13	0.54	9.15	−0.97	5.30 (100)	6.07 (100)
	2t	127	7.21	0.54	8.07	−0.96	4.15 (78)	4.96 (82)
	4t	100	5.72	0.55	7.03	−0.89	3.87 (73)	3.87 (64)
	1y	148	6.61	0.51	8.20	−0.98	4.87 (92)	5.32 (88)
4	Pre	150	9.43	0.22	5.55	−0.93	3.45 (100)	3.76 (100)
	2t	122	9.39	0.25	5.66	−0.95	3.33 (96)	3.63 (97)
	4t	105	8.11	0.32	5.83	−0.92	3.17 (92)	3.17 (84)
	1y	138	8.61	0.26	5.71	−0.84	3.44 (100)	3.58 (95)
5	Pre	156	7.42	0.54	7.79	−0.92	3.78 (100)	4.40 (100)
	2t	103	7.15	0.52	7.40	−0.95	3.65 (97)	4.12 (93)
	4t	109	6.26	0.43	6.63	−0.80	3.96 (105)	3.96 (90)
	1y	138	7.29	0.46	6.56	−0.93	3.22 (85)	3.69 (84)
6	Pre	122	5.67	0.66	7.75	−0.95	4.03 (100)	5.35 (100)
	2t	92	5.62	0.59	6.70	−0.92	3.37 (84)	4.53 (85)
	4t	97	4.95	0.55	6.02	−0.91	3.31 (82)	4.00 (75)
	½ y	119	3.65	0.79	6.41	−0.90	3.51 (87)	3.50 (65)‡

*Pt = patient; Pre = before chemotherapy; 2t = after two treatments with bleomycin; 4t = after four treatments with bleomycin; 1y = 1 year after last (fourth) treatment with bleomycin; a and b are linear regression coefficients, and r is the correlation coefficient; %Pretreatment = percentage of pretreatment value; ½ y = 6 months after last (fourth) treatment with bleomycin.

†Lung volume is TLC after four bleomycin courses.

‡Smallest lung volume is TLC after 6 months, and patient died before the 1-year stage.

DLCO/VA vs VA Relationships

A typical example of the relationships between the diffusion indexes and VA before and after four treatments with bleomycin (solid lines) and during recovery (dotted lines) is given in Figure 1 for a patient in which DLCO/VA, DLCO, and TLC were decreased due to chemotherapy. The dashed lines with shaded areas in Figure 1 represent the volume-dependent predicted values ± 1 SD.[1] The DLCO/VA vs VA relationship before treatment was within 1 SD from the predicting equation. The DLCO/VA vs VA relationship after four treatments decreased > 4 SD in parallel to the predicted and pretreatment DLCO/VA vs VA relationships. After 6 months, the DLCO/VA vs VA relationship returned toward the initial position, but remained about 2 SD below predicted and did not improve further in the next 6 months. If we regard the initial value of DLCO/VA at the initial TLC as 100% (A), then DLCO/VA at the treatment limited TLC after four treatments with bleomycin (B) decreased 25% (B with respect to A). When DLCO/VA after four treatments was compared with pretreatment DLCO/VA at a volume similar to the treatment-limited TLC (C), a larger decrease of 36% (B with respect to C) was found.

In all patients, DLCO/VA vs VA was linear before, during, and after treatment (Tables 1, 2). The slope (b) of the DLCO/VA vs VA relationships did not change after four chemotherapeutic treatments ($p = 0.30$). The relationships of DLCO/VA vs VA shifted downwards, implying that DLCO/VA at all lung volumes, including TLC, was significantly decreased with respect to its pretreatment values (p value < 0.001 for both groups).

In Tables 1, 2, we compared DLCO/VA at actual TLC during and after chemotherapy, both with the pretreatment DLCO/VA at pretreatment TLC, and at a similar lung volume equal to the TLC after four treatments with bleomycin. In patients who developed a volume restriction (Table 1), DLCO/VA at actual TLC decreased more when compared to pretreatment DLCO/VA at the same lung volume (23 ± 9%) than when compared to

Table 2—*Change in Hb Concentration, TLC, and DLCO/VA vs VA Relationship Dependent on the Stage of the Chemotherapy in the Patients Who Did Not Develop a Restrictive Ventilatory Defect

Pt	After Bleomycin Treatment and Recovery	Hb, g/L	TLC, L	DLCO/VA = a − b VA			DLCO/VA, mL CO/min/mm (% Pretreatment) Hg/L	
				b	a	r	At Actual TLC	At Similar Lung Volume
7	Pre	164	6.96	0.37	7.48	−0.97	4.89 (100)	4.98 (100)
	2t	126	7.12	0.46	7.38	−0.95	4.12 (84)	4.30 (86)
	4t	129	6.74	0.44	7.61	−0.98	4.65 (95)	4.65 (94)
	1y	161	6.44	0.31	6.62	−0.88	4.60 (94)	4.51 (91)
8	Pre	147	8.46	0.38	6.42	−0.96	3.20 (100)	3.42 (100)
	2t	137	8.23	0.37	6.07	−0.93	3.01 (94)	3.13 (92)
	4t	109	7.91	0.38	5.72	−0.93	2.76 (86)	2.76 (81)
	1y	166	7.62	0.38	6.11	−0.98	3.20 (100)	3.10 (91)
9	Pre	158	6.88	0.79	10.01	−0.96	4.58 (100)	4.67 (100)
	2t	147	6.90	0.89	9.55	−0.96	3.42 (75)	3.54 (76)
	4t	95	6.76	0.86	9.39	−0.91	3.60 (79)	3.60 (77)
	1y	142	6.49	1.03	10.85	−0.99	4.13 (90)	3.87 (83)
10	Pre	118	7.40	0.50	7.36	−0.91	3.63 (100)	3.47 (100)
	2t	103	8.09	0.53	7.46	−0.88	3.19 (88)	3.40 (98)
	4t	95	7.70	0.52	7.58	−0.88	3.58 (99)	3.58 (103)
	1y	138	8.10	0.55	7.87	−0.93	3.37 (93)	3.60 (104)
11	Pre	158	7.89	0.77	10.20	−0.96	4.15 (100)	4.26 (100)
	2t	129	8.59	0.69	9.37	−0.93	3.49 (84)	4.06 (95)
	4t	126	7.75	0.75	9.26	−0.91	3.45 (83)	3.45 (81)
	1y	156	7.94	0.72	9.85	−0.97	4.17 (101)	4.31 (101)
12	Pre	130	6.30	0.54	6.90	−0.91	3.47 (100)	3.26 (100)
	2t	105	6.62	0.50	6.09	−0.92	2.79 (80)	2.77 (85)
	4t	89	6.67	0.43	5.81	−0.91	2.92 (84)	2.92 (90)
	1y	137	6.79	0.36	5.94	−0.86	3.51 (101)	3.54 (109)
13	Pre	147	7.73	0.26	6.54	−0.91	4.51 (100)	4.53 (100)
	2t	134	7.55	0.21	5.56	−0.77	3.97 (88)	3.94 (87)
	4t	113	7.66	0.47	6.83	−0.94	3.19 (71)	3.19 (70)
	1y	140	7.27	0.51	7.55	−0.94	3.79 (84)	3.60 (80)

*See Table 1 for abbreviations.

pretreatment DLCO/VA at pretreatment TLC (the difference of both methods of comparison is 10 ± 3%; p value < 0.001, paired t test). In patients without a volume restriction (Table 2), actual DLCO/VA was compared with pretreatment DLCO/VA at pretreatment TLC. After four courses, the decrease in DLCO/VA was on average 15 ± 11% of the pretreatment value.

Discussion

Ventilation distribution was evaluated on the basis of the ratio between TLCsb and TLCmb. In all patients at all stages of treatment, the TLCsb/TLCmb ratio was > 0.85, which has been proposed as an indication for normal ventilation distribution.[22] When ventilation unequality is increased (for instance in elderly or COPD patients), this ratio is decreased and gas transfer is primarily studied in the best ventilated areas.

During bleomycin treatment, the patients developed a volume restriction or a diffusion disorder, or both. Furthermore, the relationships between DLCO or DLCO/VA and VA remained parabolic and linear, respectively, the latter with a negative slope, as in healthy volunteers. Probably because the alveolar membrane expands in a similar way, its change in gas transfer capacity is comparable.[11,12] Both relationships decreased to a lower level, partly due to a decrease in Hb concentration. To evaluate the diffusion disorder, we corrected the data to pretreatment Hb concentrations.

The data are in agreement with our hypothesis that DLCO/VA is decreased by the diffusion disorder and increased by the volume restriction. Extrapolation of DLCO/VA to patients' TLC before treatment, or to reference TLC, will result in a DLCO/VA lower than the actual DLCO/VA at actual TLC (except when no volume restriction would have occurred). Consequently, in those patients who developed a volume restriction (Table 1), the difference between DLCO/VA at actual TLC dur-

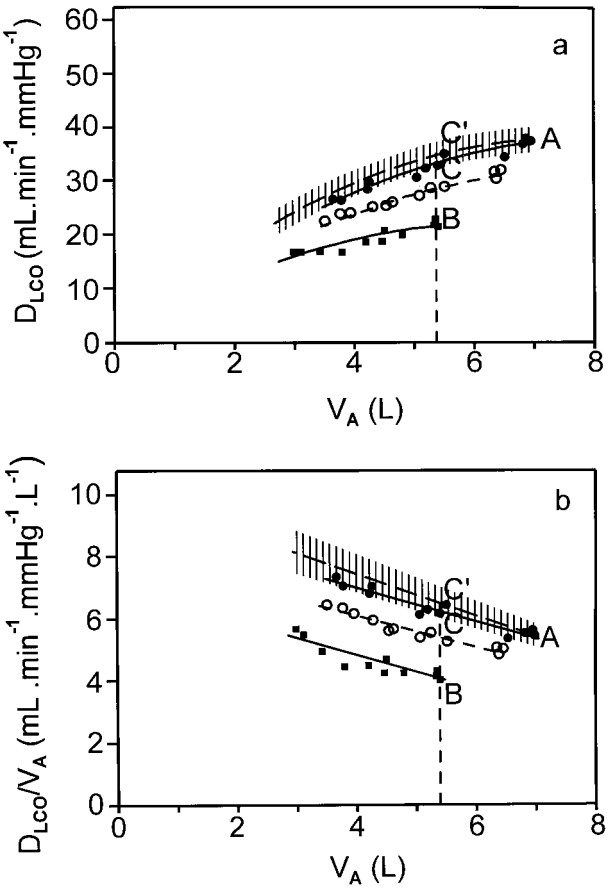

FIGURE 1. Individual example (subject 3; Table 1) of DLCO vs VA (*top, a*) and DLCO/VA vs VA relationships (*bottom, b*), respectively, before and after four courses of chemotherapy containing bleomycin (solid lines) and after 1 year of recovery (dashed lines) in a patient who developed a diffusion disorder as well as a volume restriction. The dashed lines in the shaded areas represent the volume-dependent predicted values ± 1 SD, as found in a previous study.[1] ● = before chemotherapy; ■ = after four courses with bleomycin-containing chemotherapy; ○ = 1 year after the last treatment; A = DLCO/VA and DLCO values at initial TLC; B = DLCO/VA and DLCO values at the disease-limited TLC after four courses with bleomycin-containing chemotherapy; C = DLCO/VA and DLCO values before chemotherapy at a VA equal to the TLC after chemotherapy; C' = DLCO/VA and DLCO predicted values at a VA equal to the TLC after chemotherapy.

ing chemotherapy (B in Fig 1, *bottom, b*) and the pretreatment DLCO/VA at pretreatment TLC (A in Fig 1, *bottom, b*) is significantly smaller than the difference in DLCO/VA with respect to pretreatment DLCO/VA (C in Fig 1, *bottom, b*) at similar lung volume. We concluded that comparison of DLCO/VA at a treatment-limited TLC with the DLCO/VA at the initial or predicted TLC implies an underestimation of the diffusion disorder. Since our results (Fig 1, *bottom, b*, C and C') revealed that the pretreatment values of our patients were similar to their predicted values, similar studies on other restrictive diseases can be done by comparison of the actual DLCO/VA vs VA relationship with the predicted relationship.[1]

This study supports the hypothesis that the effect of a volume restriction caused by bleomycin treatment on the diffusion indexes is comparable to that of a voluntary volume reduction in healthy volunteers. Although our results only apply for patients with a restrictive disease due to bleomycin treatment, we would recommend, at this stage of knowledge, comparing actual DLCO/VA with its predicted value at the same lung volume as the disease-limited TLC in patients with other restrictive lung diseases.[1] However, to evaluate the individual decrease in total diffusing capacity, we recommend comparing the Hb-corrected DLCO in patients with a restrictive lung disease with a predicted DLCO at predicted TLC. DLCO in percentage of its predicted value at predicted TLC reflects the total effect of both volume restriction and alveolar capillary diffusion disorder on DLCO.

In this study, we performed 12 measurements on each patient before, during, and after the treatment with bleomycin, to illustrate that the slopes of the linear DLCO/VA vs VA relationships are not changed. In practice, however, to monitor the side effect of bleomycin on lung tissue, using the estimation of DLCO and DLCO/VA at a large number of VA levels before, during, and after the courses of chemotherapy is time consuming. Therefore, we recommend determining the relationship between DLCO/VA and VA before chemotherapy and to estimate DLCO/VA during the courses of medication at the actual TLC only, or to compare DLCO/VA during chemotherapy with predicted values, taking a volume restriction into account.[1] During chemotherapy, DLCO/VA is measured at the treatment-limited TLC only and is compared with the pretreatment or predicted DLCO/VA at the same volume. To determine the relationship between DLCO/VA and VA in this study, we performed a relatively large number of measurements; in practice, about six measurements at various VA levels appear to be sufficient for proper regression analysis.

In this study, all patients were relatively young, and in normal subjects, as described in our former article,[1] the slope of the DLCO/VA vs VA relationship is steepest at younger ages. We assume that a comparable age dependency holds in patients with a restrictive lung disease. Therefore, we would postulate that the influence of volume changes on DLCO/VA is less important in elderly patients with a restrictive lung disease.

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